

SUPPORT FOR THE AMENDMENTS

Newly-added Claims 34-52 are supported by the specification at pages 4-32 and the original claims. No new matter is believed to have been added to the present application by the amendments submitted above.

REMARKS

Claims 34-52 are now pending.

Applicants would like to thank Examiner Fronda for the helpful and courteous discussion held with their representatives on February 7, 2006. During the discussions, Claim 34 presented above was discussed with respect to the outstanding rejections. The following remarks expand on the discussion with the Examiner.

The present invention relates to a microorganism belonging to enterobacteria selected from the group consisting of the genus *Enterobacter*, *Pantoea*, *Klebsiella*, *Erwinia* and *Serratia* and having L-glutamic acid productivity which is transformed by a polynucleotide sequence encoding a citrate synthase obtained from *Corynebacterium glutamicum* or *Brevibacterium lactofermentum*,

where the transformed microorganism has enhanced L-glutamic acid productivity as compared to the untransformed microorganism.

See Claim 34.

The present invention also relates to 49. (New) A process for producing L-glutamic acid, comprising:

isolating a polynucleotide sequence encoding a citrate synthase obtained from a coryneform bacterium, wherein the polynucleotide is obtainable by the polymerase chain reaction using oligonucleotide primers of SEQ ID NO: 1 and SEQ ID NO: 2;

transforming an enterobacteria with said isolated polynucleotide;

culturing said enterobacteria in a liquid medium to produce and accumulate the L-glutamic acid, wherein the transformed enterobacteria has enhanced L-glutamic acid productivity as compared to the untransformed enterobacteria; and

collecting the L-glutamic acid produced.

See Claim 49.

The rejection under 35 U.S.C. §112, first paragraph, is believed to be obviated by the amendments submitted above.

At the outset, Applicants note that the sequence of the polynucleotide sequence is known in the art. See Eikmanns et al. In the recent Capon decision, the Federal Circuit held that there is no rule that the specification must provide a specific sequence if that sequence is known in the art. A copy of the Capon decision is enclosed for the Examiner's convenience. See also the BLASTIN report enclosed with the Amendment submitted on March 7, 2005.

In view of the foregoing, the claims satisfy the written description requirement. Accordingly, withdrawal of this ground of rejection is respectfully requested.

The rejection of the claims under 35 U.S.C. §103(a) over Skillman et al. in view of Eikmanns et al. is respectfully traversed. Those references fail to suggest the claimed microorganism.

Skillman et al. disclose transforming *Enterobacter agglomerans* and *E. coli* with a gene encoding green fluorescent protein in order to examine the interactions between bacterial species and their effects on biofilm development. See the Abstract.

Eikmanns et al. disclose the sequence of a gene encoding citrate synthase from *Corynebacterium glutamicum*. See the Abstract.

One with the cited references in hand would not be motivated to transform the bacteria described by Skillman et al. with the citrate synthase gene disclosed by Eikmanns et al. This is because the purpose of the transformation described by Skillman et al is to examine the interactions between bacterial species and their effects on biofilm development. As described by Eikmanns et al., citrate synthase is the catalyst of the initial reaction of the citric acid cycle, which is not recognized to be directly involved in the interactions between bacteria and the formation of biofilms. Therefore, there is simply no motivation to transform

the bacteria described by Skillman et al. with the citrate synthase gene disclosed by Eikmanns et al.

In addition, the specification of the present application describes that the claimed invention was developed as follows:

The inventors of the present invention have bred enterobacteria by introducing a gene to improve their productivity of L-glutamic acid. Generally, it has been considered that a better effect is obtained by using a endogenous gene of a host or a gene derived from a microorganism which is a relative of the host than by introducing a heterogeneous gene when the host has a target gene for the breeding of a microorganism by gene amplification. However, the inventors of the present invention have found it much more effective, for enterobacteria, in improving the L-glutamic acid productivity of a microorganism to introduce a CS [i.e., citrate synthase] gene derived from a coryneform bacterium than to introduce a CS gene derived from a microorganism of the same species as the enterobacteria. The present invention has been accomplished based on this finding. [Paragraph bridging pages 4-5 of the specification.]

Thus, the Inventors have discovered that by going against their expectation by using a heterologous polynucleotide instead of an endogenous sequence, enhanced L-glutamic acid productivity is obtained. This is shown in the data presented in Table 1 at page 29 of the specification, which is reproduced below for convenience.

The bacterial strains listed in Table 1 are summarized below:

<u>Bacterial Strain</u>	<u>Description</u>
AJ13355	Non-transformed <i>Enterobacter agglomerans</i> (i.e., control)
AJ13355/pMWC	<i>Enterobacter agglomerans</i> transformed with citrate synthase from <i>E. coli</i> (i.e., endogenous polynucleotide)
AJ13355/pMWCB	<i>Enterobacter agglomerans</i> transformed with citrate synthase From <i>Brevibacterium lactofermentum</i> (i.e., heterologous polynucleotide)
AJ13355	Non-transformed <i>Klebsiella planticola</i> (i.e., control)
AJ13355/pMWC	<i>Klebsiella planticola</i> transformed with transformed with citrate synthase from <i>E. coli</i> (i.e., endogenous polynucleotide)
AJ13355/pMWCB	<i>Klebsiella planticola</i> transformed with transformed with citrate synthase from <i>Brevibacterium lactofermentum</i> (i.e., heterologous polynucleotide)

The results presented in Table 1 of the specification demonstrate that enhanced L-glutamic acid productivity is obtained using a heterologous polynucleotide (i.e., from *Brevibacterium lactofermentum*) encoding citrate synthase as compared to an endogenous sequence (i.e., from *E. coli*). As discussed above, this is contrary to the Inventors' expectations.

In view of the foregoing, the claimed microorganism is not obvious over Skillman et al. in view of Eikmanns et al. Accordingly, withdrawal of this ground of rejection is respectfully requested.

The rejection of the process claims under 35 U.S.C. §103(a) over Skillman et al. in view of Eikmanns et al. and Mikio et al. is respectfully traversed. Those references fail to suggest the claimed process.

Claim 46 specifies isolating a polynucleotide sequence encoding a citrate synthase obtained from a coryneform bacterium citrate, where the polynucleotide is obtainable by PCR

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amplification of chromosomal DNA using primers of SEQ ID NO: 1 and SEQ ID NO: 2.  
Claim 46 also specifies that the transformed enterobacteria has enhanced L-glutamic acid productivity as compared to the untransformed enterobacteria. The cited references fail to suggest such features in a process in which L-glutamic acid is isolated from a liquid medium. Accordingly, withdrawal of this ground of rejection is respectfully requested.

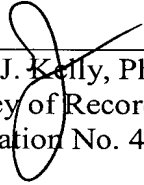
Applicants submit that the present application is in condition for allowance. Early notice to this effect is earnestly solicited.

Respectfully submitted,

OBLON, SPIVAK, McCLELLAND,  
MAIER & NEUSTADT, P.C.

Customer Number  
**22850**

Tel: (703) 413-3000  
Fax: (703) 413 -2220  
(OSMMN 06/04)

  
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James J. Kelly, Ph.D.  
Attorney of Record  
Registration No. 41,504